fore mentioned basic ideas available to those skilled in the art without departing from the spirit of the invention. The scope of this method and apparatus to detect microbes includes utilization of simultaneous excitation of multiple intrinsic microbial fluorophores with subsequent analysis of the detected emissions with methods that concurrently account for background signals and require said signals to lie within physiological ranges. All variations, modifications and configurations are intended to be within the scope of the present invention as defined in the appended claims.

WHAT IS CLAIMED:

1. A method for the detection of microbes comprising:

   a. exciting at least one intrinsic microbial fluorophore having a specific range of electromagnetic radiation wavelength above 200 nm; whereby said microbe containing intrinsic fluorophores is excited to emit fluorescence, and;

   b. detecting the fluorescence signals associated with the minima and maxima of the excited microbial fluorophores, and;

   c. subtracting the reflected/scattered excitation energies and background from the detected signals of the excited microbial fluoresence; whereby the enumeration of microbes is determined by the magnitude of the detected fluorescence.

2. The method as set forth in Claim 1, wherein the relative ratios of multiple detected, background-corrected signals are determined; whereby the detection of the physiological state of the microbes depends upon the requirement that the ratios of the background-corrected fluorescence signals lie within specified physiological ranges and that the enumeration of the microbes is determined by

the magnitude of said detected signals whose relative ratios lie within said expected ranges.

3. A method as set forth in Claim 1, wherein said microbe fluorephores are selected from the group consisting of nucleic acid polymers, tryptophan-containing proteins, tyrosine-containing proteins, adenosine triphosphate, calcium dipicinolate, reduced pyridine nucleotides, flavins, porphyrin-containing proteins, and other components excited in the 610-670 nm region.

4. The method of Claim 1 wherein the viable microbes to be detected include bacteria, fungi, protazoa, and rickettsiae; and the intrinsic microbial fluorophores used to detect the microbes include nucleic acid polymers, tyrosine-containing proteins, tryptophan-containing proteins, adenosine triphosphate, reduced pyridine nucleotides, flavins, porphyrin-containing proteins, and others excited in the 610-670 nm region.

5. The method of Claim 1 wherein non-viable microbes to be detected include bacteria, fungi, protazoa, and rickettsiae; and the intrinsic microbial fluorophores used to detect the microbes include nucleic acid polymers, tryptophan-containing proteins, tyrosine-containing proteins, reduced pyridine nucleotides, flavins, porphyrin-containing proteins, and others excited in the 610-670 nm region.

6. The method of Claim 1 wherein the microbes to be detected are bacterial endospores and the intrinsic fluorophores used to detect the endospores include nucleic acid polymers, tyrosine-containing proteins, tryptophan-containing proteins, calcium dipicolinic acid, and others excited in the 610-670 nm region.

7. The method of claim 1 wherein the microbes to be detected include viruses, and the intrinsic fluorophores used to detect the viruses include nucleic acid polymers, tyrosine-containing proteins and tryptophan-containing proteins.

8. A method for the detection of microbes comprising:

a. excitation of multiple intrinsic microbial fluorophores with ultraviolet electromagnetic radiation having wavelengths between 200 and 300 nm, whereby any microbes present containing intrinsic fluorophores are excited to emit fluorescence, some of whose fluorescence is self-absorbed to excite other microbial fluorophores that in turn emit fluorescence;

b. detecting the fluorescence signals associated with the minima and maxima of the excited microbial fluorophores;

c. subtracting the fluorescence from the reflected scattered excitation energies and background from the detected signals; and

d. determining that the relative ratios of the detected, background-corrected signals lie within physiological ranges, whereby the enumeration of the microbes is determined by the magnitude of the detected signals whose relative ratios lie within physiological ranges.

9. A method as set forth in Claim 8, wherein the intrinsic microbial fluorophores of said microbes include one or more of the group consisting of nucleic acid polymers, tryptophan-containing proteins, adenosine triphosphate and calcium dipicinolate compounds.

10. A method as set forth in Claim 8, wherein secondary-excited microbial fluorophores include one or more of the group consisting of calcium dipicinolate,

reduced pyridine nucleotides, flavins, porphyrin-containing proteins, cellular components excited in the 610-670 nm region, and the like.

11. The method of Claim 8 wherein the viable microbes to be detected include bacteria, fungi, protazoa, and rickettsiae and the intrinsic microbial fluorophores used to detect the microbes are selected from the group consisting of nucleic acid polymers, tyrosine-containing proteins, tryptophan-containing proteins, adenosine triphosphate, reduced pyridine nucleotides, flavins, porphyrin-containing proteins, and cellular components excited in the 610-670 nm region.

12. The method of Claim 8 wherein the non-viable microbes to be detected include bacteria, fungi, protazoa, and rickettsiae; and the intrinsic microbial fluorophores used to detect the microbes are selected from the group consisting of nucleic acid polymers, tyrosine-containing proteins, tryptophan-containing proteins, reduced pyridine nucleotides, flavins, porphyrin-containing proteins, and cellular components excited in the 610-670 nm region.

13. The method of Claim 8 wherein the microbes to be detected are bacterial spores and the intrinsic fluorophores used to detect the spores include nucleic acid polymers, tyrosine-containing proteins, tryptophan-containing proteins, calcium dipicolinic acid, and spore components excited in the 610-670 nm region.

14. Apparatus for the detection of microbes on a non-living surface or in air or liquid comprising:

a. means for directing electromagnetic radiation towards the sample, said means adapted to emit radiation energies capable of exciting at least one intrinsic microbial fluorophore;

b. at least one detector for electromagnetic radiation capable of converting the emitted, or reflected scattered radiation into electrical signals, said detector adapted to detect electromagnetic radiation at wavelengths above 320 nm to detect the minima and maxima associated with the fluorescence emission of said microbial fluorophores; and

c. means for analyzing the electrical signals corresponding to the fluorescence of the intrinsic microbial fluorophores, and the reflected scattered excitation energies to determine the presence of microbes.

15. The method as defined in claim 1 wherein the electromagnetic waves are directed towards the microbes in time-modulated pulses.

16. The apparatus defined in Claim 14 wherein the means for directing electromagnetic radiation includes means for time-modulating the electromagnetic radiation.

17. A method for the detection of microbial proteinaceous toxins comprising:

a. exciting at least one intrinsic microbial fluorophore having a specific range of electromagnetic radiation wavelength above 200 nm; whereby said microbe present containing intrinsic fluorophores is excited to emit fluorescence, and;

b. detecting the fluorescence signals associated with the minima and maxima of the excited microbial fluorophores, and;

c. subtracting the reflected scattered excitation energies and background from the detected signals of the excited microbial fluoresence; whereby enumeration of the microbial toxin is determined by the magnitude of said detected background-corrected fluorescence.

18. A method as set forth in Claim 17, wherein said microbe fluorephores are selected from the group consisting of tryptophan-containing proteins and tyrosine-containing proteins.

19. A method for the detection of non-viable bacteria and spores comprising:

a. exciting at least one intrinsic microbial fluorophore having a specific range of electromagnetic radiation having wavelengths between 550 and 700 nm; whereby said microbe present containing intrinsic fluorophores is excited to emit fluorescence, and;

b. detecting the fluorescence signals associated with the minima and maxima of the excited microbial fluorophores, and;

c. subtracting the reflected scattered excitation energies and background from the detected signals of the excited microbial fluoresence; whereby the enumeration of non-viable bacteria and spores is determined by the magnitude of the detected fluorescence.

20. The method as set forth in Claim 19, wherein the relative ratios of multiple detected, background-corrected signals are determined; whereby the distinction between spores and non-viable bacteria depends upon the requirement that the ratios of the background-corrected fluorescence signals lie within specified physiological ranges and that the enumeration of spores is determined by the magnitude of said detected signals whose relative ratios lie within said expected ranges.

21. A method of Claim 19 wherein the intrinsic microbial fluorophores used to detect the non-viable bacteria and bacterial spores are selected from a group including

flavins, porphyrin-containing proteins, other components excited in the 610-670 nm region.

22. The method of Claim 19 wherein the non-viable bacteria and bacterial spores are detected on surfaces, inside paper envelopes, through paper, in solution and in aerosols.

23. A method for the detection of spores and non-viable bacteria comprising:

a. excitation of multiple intrinsic microbial fluorophores with electromagnetic radiation having wavelengths between 550 and 640 nm, whereby any spores and non-viable bacteria present containing intrinsic fluorophores are excited to emit fluorescence, some of whose fluorescence is self-absorbed to excite other spore fluorophores that in turn emit fluorescence;

b. detecting the fluorescence signals associated with the minima and maxima of the excited microbial fluorophores;

c. subtracting the fluorescence from the reflected/scattered excitation energies and background from the detected signals; and

d. determining that the relative ratios of the detected, background-corrected signals lie within physiological ranges, whereby the enumeration of the non-viable bacteria and bacterial spores is determined by the magnitude of the detected signals whose relative ratios lie within physiological ranges.

24. A method as set forth in Claim 23, wherein the intrinsic microbial fluorophores of said microbes include one or more of the group consisting of flavins, porphyrin-containing proteins, and other components excited in the 610-670 nm region.

25. A method as set forth in Claim 23, wherein secondary-excited microbial fluorophores include one or more of the group consisting of intrinsic components excited in the 610-680 nm region, and the like.

26. The method of Claim 23 wherein the non-viable bacteria and spores are detected on surfaces, inside paper envelopes, through paper, in solution and in aerosols.

27. A method as set forth in Claim 23, wherein the spores and non-viable bacteria are detected inside paper envelopes and the secondary-excited microbial fluorophores are excited by emissions from the excited paper products.